

## REMARKS

By a non-final Office Action dated July 21, 2005, the Examiner in charge of this case rejected the claims of this application on a variety of grounds. The disposition of the claims is as follows: Claims 1-4, 6-10, 12, 13, 15-19, 21, 22 and 24-32 are currently pending in the application; Claims 28-32 have been withdrawn from consideration as being drawn to a non-elected invention; and Claims 1-4, 6-10, 12, 13, 15-19, 21, 22 and 24-27 are rejected. The applicants have responded to the rejections by submitting the amendments and comments set forth hereinbelow. Based on this submission, reconsideration of the merits of this patent application is respectfully requested.

At the outset, in responding to the Examiner's rejections applicants submit that the format of independent Claims 1 and 10 have been amended to more clearly layout the steps needed for effectively practicing the methods of the invention. These claims now include two steps: (a) introducing the trifolitoxin-producing bacterium in the plant and (b) observing control of crown gall disease on the plant susceptible to the disease, compared to a plant not exposed to the trifolitoxin-producing bacterium. Support for such an amendment may be found, for example, at pg. 3, ¶ 7; pg. 5, ¶ 20; pg. 9, ¶ 33; pg. 9, ¶ 34; and pg. 13, ¶¶ 43 and 46 of the specification.

In changing the format of these claims, applicants have also amended Claim 1 and 10 to further clarify that the  $\alpha$ -proteobacteria enters the stem of the plant through a wound site on the plant. Support for such an amendment may be found, for example, at pg. 2, ¶ 7; and pgs. 14-16, ¶¶ 49-59 of the specification. Also, although this trifolitoxin-producing bacterium may be applied to and effective for controlling crown gall disease on all plant species susceptible to it, in an effort to improve clarity of the claims, applicants have amended Claims 1 and 10 to include grape plants, fruit trees and rose plants. Accordingly, dependent Claims 8 and 17 identifying the specific plant types have been cancelled herein.

### Claim Objections – 35 U.S.C. §112; Second Paragraph

With respect to the Examiner's objection to Claims 12 and 13 under 35 U.S.C. §112; 2<sup>nd</sup> Paragraph, applicants respond by correcting the dependency of Claim 12.

### Claim Objections – 35 U.S.C. §112; First Paragraph

Claims 1-4, 6-10, 12-13, 15-19, 21, 22, and 24-27 stand rejected under 35 U.S.C. 112, 1<sup>st</sup> paragraph, as failing to comply with the enablement requirement as set forth on pages 6-9

of the Office Action mailed February 18, 2004. The Examiner acknowledges the submission of the Declaration from Eric Triplett, but maintains the assertion that the terms “ $\alpha$ -proteobacteria” and “plants” in the claims are overly broad in scope. Specifically, the Examiner asserts that the Declaration gives no evidence on hosts plants other than Nicotiana and Grape, or for other strains of Agrobacterium not adapted to particular plant host species.

In response, applicants maintain that the term “plant” has already been limited to plant species that are susceptible to crown gall disease. As indicated in Dr. Triplett’s Declaration, taxonomically divergent plant species are responsive to the biological control of crown gall via various strains of  $\alpha$ -proteobacteria. Although, applicants have supplied only evidence of effectively controlling crown gall on Nicotiana and grapes, using the trifolitoxin-producing bacterium there are a number of indicators that would predict successful use of the bacterium to control galling in other plant varieties. The applicants believe that the inventive method is applicable to all plants because, for example, crown gall disease occurs when the soil bacterium ( $\alpha$ -proteobacterium) enters the stem of the plant through a wound site. Thus, providing that the crown-gall susceptible plant is wounded and capable of being infected the required level of enablement has been provided for one of ordinary skill to practice the invention, i.e., applying the trifolitoxin-producing bacterium onto the wounded stem and observing subsequent control of crown gall disease. Nevertheless, to obviate the issue without the need for further discussion, as indicated above, Claims 1 and 10 have been amended to include varieties of grapes, fruits and roses.

Furthermore, with respect to the term “ $\alpha$ -proteobacteria,” applicants maintain that the scope is not broad because it has been limited to Rhizobiaceae bacteria which (1) have been genetically engineered to express a *txf* operon, (2) produce trifolitoxin, and (3) control crown gall disease. Again, however, to clarify the claim and obviate the issue, as indicated above, Claims 1 and 10 have been amended to indicate that the trifolitoxin-producing  $\alpha$ -proteobacteria enter the stem of the plant through a wound site on the plant. Therefore, applicants respectfully request that in view of these claim amendments, the rejection be reconsidered and withdrawn.

#### Claim Rejections – 35 U.S.C. §102

Claims 1, 7, 9, 10, 16, 18 and 26 stand rejected under 35 U.S.C. 102(b) as being anticipated by Robleto, et al. *Environmental Microbiology*, (1998), Vol. 64, No. 7, page 2630-2633. The Examiner alleges that the Robleto teaches a method of controlling crown

gall on plants by introducing on to the plant an effective amount of biologically pure culture of an  $\alpha$ -proteobacteria, where the  $\alpha$ -proteobacteria is a *Rhizobium* engineered to express the trifolitoxin operon, expresses a pT2TFXK plasmid, produces trifolitoxin and where the plant is a seed.

In response, applicants submit that Robleto discloses a genetically engineered method to improve nodulation competitiveness in field grown plants by inoculating seeds with mixtures of *Rhizobium* strains at different ratios (see Abstract). The experimental results showed that addition of the TFX production phenotype significantly increased nodule occupancy under field conditions. Therefore, the authors concluded that the over production of trifolitoxin conferred by pT2TFXK greatly expanded the range of species of root nodule bacteria inhibited by trifolitoxin.

Applicants submit that there are several key differences between the disclosure of Robleto and the present invention. Robleto's method of increasing nodule competitiveness uses a different site of infection and a different mechanism of infection. For example, *Rhizobia* infect root hairs; in contrast *Agrobacteria* infect wounded or de-differentiated (e.g., callus culture) plant tissue. Robleto's method also uses a different mechanism of infection. For example, it is well known in the art that antibiotics that are effective in soil or *in vitro* may not be effective or involved in controlling disease. (See, for example, Burr et al., "Biological control of grape crown gall with non-tumorigenic *Agrobacterium vitis* strain F2/5," Am. J. Enol. Vitic (1993) 45:213-219; which describes an antibiotic produced by *Agrobacterium vitis* F2/5, which does NOT produce any disease control phenotype.) Thus, Robleto's method for nodulation improvement would not be equally effective for controlling crown gall disease because as indicated by Dr. Triplett's Declaration, nodulation and galling are two different processes which occur in very different environments.

Furthermore, applicants submit that in view of the claim amendments set forth hereinabove, Robleto does not teach all of the method steps (i.e., introducing trifolitoxin-producing bacteria on a crown gall susceptible plant and observing control of crown gall disease) or the starting materials of the claimed processes. Accordingly, applicants believe that the method disclosed in Robleto does not anticipate the claimed method.

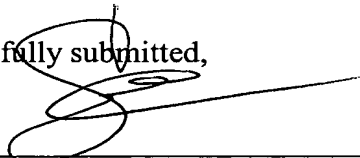
Claims 1, 6, 7, 9, 10, 15, 16, 18 and 26 also stand rejected under 35 U.S.C. 102(b) as being anticipated by Robleto, as evidence by Breil, et al., *J. Bacteriol.* (1993), vol. 175, pages 3696-3702 and Breil et al., NCBI Accession No. L06719, locus RHMTFXA2G, August 4, 1993.

In response, applicants submit that Breil discloses DNA sequence analysis, subcloning and insertion mutation analysis of gene regions required for TFX production. In view of the foregoing claim amendments and remarks it is believed that neither Robleto and/or Breil alone or in combination make any sort of connection between TFX- producing bacteria and the biological control of crown gall disease. Thus, Breil does not cure the deficiencies of Robleto.

Accordingly, in view of the above claim amendments and remarks, the application is now believed to be in condition for allowance. Applicants respectfully request that a timely Notice of Allowance be issued in this case.

A petition for extension of time for one month is enclosed so this response will be considered as timely filed. Please charge the fee to Deposit Account No. 17-0055. No other fee is believed to be due in connection with this response. However, if any other fee is due in this or any subsequent response, please charge the fee to the same Deposit Account No. 17-0055.

Respectfully submitted,



---

Sara D. Vinarov, Reg. No.: 48,524  
Attorney for Applicants  
QUARLES & BRADY LLP  
P.O. Box 2113  
Madison, WI 53701

TEL 608/251-5000  
FAX 608/251-9166